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Mazdoor Kisan Shakti Sangathan

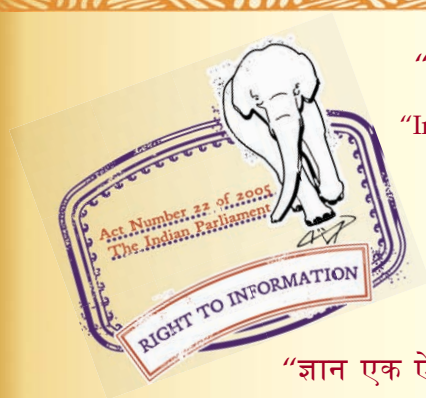
“The Right to Information, The Right to Live”

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“Step Out From the Old to the New”

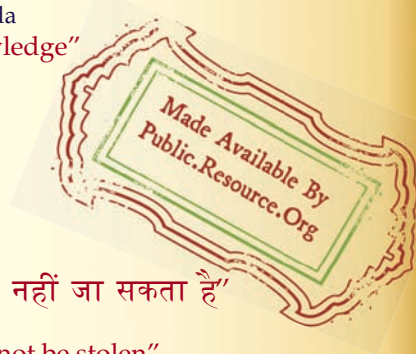
IS 11062 (1984): Method for estimation of total dietary fibre in foodstuffs [FAD 16: Foodgrains, Starches and Ready to Eat Foods]



“ज्ञान से एक नये भारत का निर्माण”

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“ज्ञान एक ऐसा खजाना है जो कभी चुराया नहीं जा सकता है”

Bhartrhari—Nitiśatakam

“Knowledge is such a treasure which cannot be stolen”

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IS : 11062 - 1984

Indian Standard

METHOD FOR ESTIMATION OF
TOTAL DIETARY FIBRE IN FOODSTUFFS

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INDIAN STANDARDS INSTITUTION
MANAK BHAVAN, 9 BAHADUR SHAH ZAFAR MARG
NEW DELHI 110002

Indian Standard

METHOD FOR ESTIMATION OF TOTAL DIETARY FIBRE IN FOODSTUFFS

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Indian Standard

METHOD FOR ESTIMATION OF TOTAL DIETARY FIBRE IN FOODSTUFFS

0. FOREWORD

0.1 This Indian standard was adopted by the Indian Standards Institution on 20 September 1984, after the draft finalized by the Food Hygiene Sectional Committee had been approved by the Agricultural and Food Products Division Council.

0.2 Dietary fibres (DF) are polysaccharides and lignin present in the human diet which are not digested by the endogenous secretions of the upper gastro-intestinal tracts. Dietary fibre may also include other non-digestible polysaccharides, such as gums and mucilages as well as other plant cell components, for example, cell wall, proteins, non-lignin phenols, cutin, phytic acid, ester-linked acetic acid and minerals.

0.3 Prevalance of many large bowel diseases, varicose veins, diabetes, ischemic heart disease, obesity, etc, are related to the consumption of highly refined foods having low dietary fibre content. In view of this the information on the dietary fibre content of the foods has acquired great significance in human health and diseases.

0.4 The different methods of analysis of food fibres can broadly be classified as those based on direct carbohydrate determination and on gravimetric determination. However, this standard specifies the gravimetric method only. It also includes provision for simultaneous determination (if desired), of available carbohydrates, such as starch and free sugars.

0.5 In preparation of this draft considerable assistance has been derived from the National Institute of Nutrition, Hyderabad.

0.6 In reporting the result of a test or analysis made in accordance with this standard, if the final value, observed or calculated, is to be rounded off, it shall be done in accordance with IS : 2-1960*. The number of significant places retained in the rounded off value should be the same as that of the specified value in this standard.

*Rules for rounding off numerical values (revised).

1. SCOPE

1.1 This standard specifies a gravimetric method for the determination of total dietary fibre after the enzymatic digestion of starch and protein in foodstuffs.

2. PRINCIPLE

2.1 The fat and moisture free sample of food is enzymatically digested to solubilize starch and protein and the residue remained behind is estimated gravimetrically as insoluble fraction of food fibre. The soluble fibre complex is precipitated from the enzyme digest and separated. The total dietary fibre content of the food sample is the sum of the insoluble and soluble fractions. This value is expressed as grams per 100 g of the dry weight of the original foodstuff.

3. APPARATUS

3.1 Autoclave

3.2 Centrifuge — with an angular head and having a capacity to accommodate six centrifuge tubes.

3.3 Centrifuge Tubes — conical, heat resistant, 100 ml capacity.

3.4 Incubator — maintained at $37 \pm 1^\circ\text{C}$.

3.5 Vacuum Oven — or hot air oven.

4. REAGENTS

4.1 Quality of Reagents — Unless specified otherwise, pure chemicals shall be employed in tests and distilled water (see IS : 1070-1977*) shall be used when the use of water as a reagent is intended.

NOTE — 'Pure chemicals' shall mean chemicals that do not contain impurities which affect the result of analysis.

4.2 Acetone

4.3 Chloroform — Methanol mixture — 2 : 1 v/v.

4.4 Ethyl Alcohol — 80 percent v/v.

4.5 Glucoamylase — of *Rhizopus* mold origin, having an activity of 5 000 — 10 000 units per gm solid (see Note 1 under 4.12).

4.6 Hydrochloric Acid — 5 N.

4.7 Pancreatin — of porcine pancreas origin, having an activity of X N F specifications (see Note 2 under 4.12).

*Specification for water for general laboratory use (second revision).

4.8 Pepsin — of porcine Stomach Mucosa origin having an activity of 30 units per mg solid.

4.9 Diethyl Ether

4.10 Phosphate Buffer — 0.1 M, pH 6.0.

4.11 Sodium Hydroxide — 3 N.

4.12 Thymol

NOTE 1 — One unit of glucoamylase liberates 1.0 mg of glucose from starch in 3 min at pH 4.5 at 55°C. Using soluble starch almost theoretical yields of glucose are obtained.

NOTE 2 — Pancreatin contains many enzymes, including amylase, trypsin, lipase, ribonuclease and protease. The National Formulary (N F) and the US Pharmacopoeia (USP) specify amylase, protease and lipase only. N F specifies that Pancreatin will convert not less than 25 times its weight of potato starch into soluble carbohydrates in 5 minutes in water at 40°C, will digest not less than 25 times its weight of casein in 60 minutes at pH 7.5 at 40°C and will release not less than 2 micro equivalents of acid per minutes per mg pancreatin from olive oil at pH 9.0 at 37°C.

5. PROCEDURE

5.1 Preparation of Assay Sample

5.1.1 The sample taken for the assay shall be representative of the entire lot. The sample of foodgrains shall be milled to a particle size of 0.5 — 1 mm in a Wiley mill. Moist foodstuffs such as fruits and vegetables shall be sliced and homogenized before drying. The dry matter content of all the samples is determined as the weight after drying for 5 hour at 105°C.

5.1.2 Extraction of Liquids — This step is recommended for foodstuffs containing more than 5 percent of lipids to facilitate the subsequent removal of free sugars by ethyl alcohol extraction. Extract the lipids by 5 hours soxhlet extraction using chloroform methanol mixture. This system removes the free, bound and polar lipids. Estimate quantitatively the lipid content of the sample, if required. Proceed with the lipid free residue for further analysis.

5.1.3 Removal of Free Sugars — Take about 3 g moisture and lipid free sample in a glass centrifuge tube of 100 ml capacity. Add 50 ml of boiling 80 percent ethyl alcohol. Stir well and centrifuge at 2000 rpm for 5 minutes. Discard the supernatant. Repeat the 80 percent ethyl alcohol treatment two more times.

NOTE — If the free sugar content of the sample is to be estimated then collect the ethyl alcohol extract quantitatively.

5.2 Estimation of Dietary Fibre

5.2.1 Pepsin Digestion — To the centrifuge tube containing the sample residue (5.1.3) add 50 ml of water and autoclave at 120°C for 20 minutes to gelatinize the starch. Cool the contents of the tube and adjust to pH 1.5 with 5M HCL Add 50 mg pepsin and 200 ul of chloroform, incubate at 37°C for 20 hours. Adjust to pH 6.0 with 3 N NaOH and proceed to the next step.

5.2.2 Pancreatin and Glucoamylase Digestion — To the contents of the centrifuge tube, add 25 ml of phosphate buffer (4.10), 100 mg of pancreatin, 20 mg of glucoamylase and a few crystals of thymol. Incubate at 37°C for 18 hours. Centrifuge at 3 000 g for 30 minutes. Collect the supernatant and wash the residue three times with water. Add the washings to the supernatant. Take the residue after washings for the estimation of insoluble fraction of the dietary fibre. Proceed for the determination of the soluble fraction taking the supernatant along with the washings.

5.2.3 Insoluble Fraction of Dietary Fibre — Wash the residue from 5.2.2 three times with acetone and then with ether. Dry the residue to a constant weight in a hot air-oven or vacuum oven. This represents the insoluble fraction of the dietary fibre.

5.2.4 Soluble Fraction of the Dietary Fibre — Dilute the supernatant from 5.2.2, with four volumes of ethyl alcohol. Centrifuge at 3 000 g for 30 minutes and collect the residue. Wash the residue, three times with ethyl alcohol, three times with acetone and then with diethyl ether. Dry to a constant weight. This represents the soluble fraction of the dietary fibre.

NOTE — If starch content of the sample is to be determined, collect the supernatant from this step quantitatively along with the washings and proceed for the estimation of hexones.

5.3 Calculations

5.3.1 Calculate the total dietary fibre in the food sample as follows :

$$\text{Total Dietary fibre} = \frac{(\text{Mass of soluble fraction} + \text{Mass of insoluble fraction}) \times 100}{\text{Mass of the sample}}$$

5.3.2 Express the results as total dietary fibre content as g/100 g on dry mass basis after correcting for the lipid content.